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# Draft Genome Sequence of the Phyllosphere Model Bacterium Pantoea agglomerans 299R

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Bacteria belonging to the genus *Pantoea* are common colonizers of plant leaf surfaces. Here, we present the draft genome sequence of *Pantoea agglomerans* 299R, a phyllosphere isolate that has become a model strain for studying the ecology of plant leaf-associated bacterial commensals.

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Copyright © 2013 Remus-Emsermann et al. This is an open-access article distributed under the terms of the Creative Commons Attribution 3.0 Unported license. Address correspondence to Johan H. J. Leveau, jleveau@ucdavis.edu.

The phyllosphere, or air-surface interface of plant leaves, provides a habitat to a large and diverse community of microorganisms, including bacteria, yeasts, and filamentous fungi (1, 2). Through interactions with each other, their host, and the atmosphere, phyllosphere microorganisms impact the health of plants, humans, and the planet, for example as foliar pathogens or disease protectants of agricultural crops (3), as enteropathogenic contaminants on fresh produce (4), or as ice-nucleating agents contributing to cloud formation (5). Exposure to UV radiation and desiccation renders the phyllosphere a hostile environment that demands great functional hardiness and plasticity from its microbial inhabitants (6).

Pantoea agglomerans 299R (Pa299R; syn. Erwinia herbicola 299R) is a spontaneous rifampin-resistant derivative of isolate 299, a pigmented bacterium that was recovered from healthy leaves of a Bartlett pear tree near Healdsburg, CA (7). As a model organism for the study of nonpathogenic bacterial epiphytes, Pa299R has contributed greatly to our understanding of phyllosphere-specific adaptations (8), patterns of bacterial aggregation and dispersion on leaves (9, 10), competition for space and nutrients (11, 12), lateral heterogeneity of the phyllosphere environment (13, 14), and impact of this heterogeneity on bacterial survival and growth (15, 16). Availability of the Pa299R genome sequence will facilitate "omics"-based studies with this model strain and accelerate the discovery of bacterial genes underlying phyllosphere fitness.

Pa299R was obtained from Steve Lindow (University of California, Berkeley). Genomic DNA was isolated from an overnight Luria-Bertani (LB) culture using a DNeasy blood and tissue kit (Qiagen, Venlo, The Netherlands) and paired-end Illumina-sequenced by BaseClear (Leiden, The Netherlands). Total RNA was isolated using RNA Protect and an RNeasy Mini Kit (Qiagen) from cultures on 0.4% glucose M9 medium with or without 0.2% Casamino Acids and sent to the UC Davis Genome Center for rRNA depletion using a Ribo-zero rRNA removal kit (Epicentre, Madison, WI), library construction, and Illumina HiSeq2000 sequencing (single reads, 50 cycles). After quality filtering, 583 Mbp of DNA and 651 Mbp of unique RNA reads were assembled, using

Ray 1.7 (17) and a 21-bp kmer size, into 109 contigs ( $\geq$ 200 bp, N<sub>50</sub> length: 109,356 bp; average length, 42,031 bp), representing 4,581,483 bp of genomic DNA (269-fold coverage) with a G+C content of 54.29%. Gene prediction by RAST (18) uncovered 4,194 coding sequences and 62 tRNAs.

The Pa299R genome revealed many adaptations consistent with an epiphytic lifestyle, including genes for high-affinity uptake and utilization of the photosynthates sucrose, fructose, and glucose, for repair of UV-damaged DNA, and for production of the osmoprotectants betaine and trehalose. Pa299R possesses a Pantoea-typical LPP-1 plasmid (19), coding for the biosynthesis of thiamine and the pigment zeaxanthin. Presence of the ipdC gene for production of the plant hormone indole 3-acetic acid (7) was confirmed, but no genes were found for synthesis of pantocin A, a Pantoea-characteristic antibiotic (20). The spontaneous resistance of Pa299R to rifampicin resulted from a D516V substitution in the rpoB gene product.

**Nucleotide sequence accession numbers.** This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession number ANKX00000000. The version described in this paper is the first version, ANKX01000000.

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This is NIOO-KNAW publication 5414. Assembly and annotation of the *Pa*299R genome sequence were performed by M.N.P.R.-E. and E.B.K. Phyllosphere-specific analysis of the genome data was done by R.T. and J.H.J.L., with contributions from M.L.M. An early draft manuscript was written by M.N.P.R.-E. as part of his PhD thesis; the submitted version was written by J.H.J.L., with inputs, edits, and final approval by all other authors.

#### **REFERENCES**

- 1. Meyer KM, Leveau JHJ. 2012. Microbiology of the phyllosphere: a playground for testing ecological concepts. Oecologia 168:621–629.
- Vorholt JA. 2012. Microbial life in the phyllosphere. Nat. Rev. Microbiol. 10:828–840.

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- 3. Newton AC, Gravouil C, Fountaine JM. 2010. Managing the ecology of foliar pathogens: ecological tolerance in crops. Ann. Appl. Biol. 157: 343–359
- 4. Brandl MT. 2006. Fitness of human enteric pathogens on plants and implications for food safety. Annu. Rev. Phytopathol. 44:367–392.
- 5. Morris CE, Sands DC, Bardin M, Jaenicke R, Vogel B, Leyrponas C, Ariya PA, Psenner R. 2011. Microbiology and atmospheric processes: research challenges concerning the impact of airborne micro-organisms on the atmosphere and climate. Biogeosciences 8:17–25.
- Leveau JHJ. 2006. Microbial communities in the phyllosphere, p. 334–367. *In* Riederer M, Müller C (ed), Annual plant reviews, vol 23. Biology of the plant cuticle. Blackwell Publishing, Oxford, United Kingdom.
- Brandl M, Clark EM, Lindow SE. 1996. Characterization of the indole-3-acetic acid (IAA) biosynthetic pathway in an epiphytic strain of *Erwinia herbicola* and IAA production *in vitro*. Can. J. Microbiol. 42:586–592.
- Brandl MT, Lindow SE. 1998. Contribution of indole-3-acetic acid production to the epiphytic fitness of *Erwinia herbicola*. Appl. Environ. Microbiol. 64:3256–3263.
- Monier JM, Lindow SE. 2005. Aggregates of resident bacteria facilitate survival of immigrant bacteria on leaf surfaces. Microb. Ecol. 49:343–352.
- Tecon R, Leveau JHJ. 2012. The mechanics of bacterial cluster formation on plant leaf surfaces as revealed by bioreporter technology. Environ. Microbiol. 14:1325–1332.
- Monier J-M, Lindow SE. 2005. Spatial organization of dual-species bacterial aggregates on leaf surfaces. Appl. Environ. Microbiol. 71: 5484-5493.
- Wilson M, Lindow SE. 1994. Coexistence among epiphytic bacterial populations mediated through nutritional resource partitioning. Appl. Environ. Microbiol. 60:4468–4477.

- 13. **Brandl MT, Quinones B, Lindow SE.** 2001. Heterogeneous transcription of an indoleacetic acid biosynthetic gene in *Erwinia herbicola* on plant surfaces. Proc. Natl. Acad. Sci. U. S. A. 98:3454–3459.
- Leveau JHJ, Lindow SE. 2001. Appetite of an epiphyte: quantitative monitoring of bacterial sugar consumption in the phyllosphere. Proc. Natl. Acad. Sci. U. S. A. 98:3446–3453.
- Remus-Emsermann MNP, Leveau JHJ. 2010. Linking environmental heterogeneity and reproductive success at single-cell resolution. ISME J. 4:215–222.
- Remus-Emsermann MNP, Tecon R, Kowalchuk GA, Leveau JHJ. 2012.
  Variation in local carrying capacity and the individual fate of bacterial colonizers in the phyllosphere. ISME J. 6:756–765.
- Boisvert S, Laviolette F, Corbeil J. 2010. Ray: simultaneous assembly of reads from a mix of high-throughput sequencing technologies. J. Comput. Biol. 17:1519–1533.
- 18. Aziz RK, Bartels D, Best AA, DeJongh M, Disz T, Edwards RA, Formsma K, Gerdes S, Glass EM, Kubal M, Meyer F, Olsen GJ, Olson R, Osterman AL, Overbeek RA, McNeil LK, Paarmann D, Paczian T, Parrello B, Pusch GD, Reich C, Stevens R, Vassieva O, Vonstein V, Wilke A, Zagnitko O. 2008. The RAST server: rapid annotations using subsystems technology. BMC Genomics 9:75.
- De Maayer P, Chan W-Y, Blom J, Venter SN, Duffy B, Smits THM, Coutinho TA. 2012. The large universal *Pantoea* plasmid LPP-1 plays a major role in biological and ecological diversification. BMC Genomics 13:625.
- Smits THM, Rezzonico F, Kamber T, Blom J, Goesmann A, Ishimaru CA, Frey JE, Stockwell VO, Duffy B. 2011. Metabolic versatility and antibacterial metabolite biosynthesis are distinguishing genomic features of the fire blight antagonist *Pantoea vagans* C9-1. PLoS One 6:e22247. doi: 10.1371/journal.pone.0022247.